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10/612,410	07/03/2003	Robert P. Bennett	IVGN 340	3567

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INVITROGEN CORPORATION
C/O INTELLEVATE
P.O. BOX 52050
MINNEAPOLIS, MN 55402

EXAMINER	
HILL, KEVIN KAI	

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1633	

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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/612,410	BENNETT, ROBERT P.	
	Examiner	Art Unit	
	Kevin K. Hill, Ph.D.	1633	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *26 February 2007*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *19-21,28-38,60-62,69-79,104-106,113-128,137 and 138* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *19-21,28-38,60-62,69-79,104-106,113-128,137 and 138* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

Detailed Action

Amendments

Applicant's amendments to Claims 19, 35, 60-61, 104-105 and 121, in the reply filed February 26, 2007, is acknowledged. Claim 61 is annotated "(Original)", but is in fact, amended. Also acknowledged are Applicant's cancellation of Claims 1-18, 22-27, 39-59, 63-68, 80-103, 107-112, 129-135 and 139-156.

1. Claims 19-21, 28-38, 60-62, 69-79, 104-106, 113-128 and 137-138 are under consideration.

Double Patenting

2. **The prior rejection of Claims 19, 28, 30, 32, 34-38, 60, 69-71, 73, 75, 104, 113-116, 118-121 and 126-128** on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claims 45-55 of U.S. Patent No. 6,720,140 **is withdrawn** in light of Applicant's amendment to base Claims 19, 60 and 104 to include the limitation of SEQ ID NO:6 which is not taught by the prior cited art.

Claim Rejections - 35 USC § 112

3. **The prior rejection of Claims 19, 35, 104 and 121 under 35 U.S.C. 112, first paragraph**, as failing to comply with the written description requirement **is withdrawn** because Applicant has deleted the phrase "mutants, variants and derivatives of the recombination sites of (a), (b), (c), (d), (e), (f), (g), (h) or (i) which retain the ability to undergo recombination" from the claims.

4. **Claims 19-38, 60-79, 104-128 and 136-138 are rejected under 35 U.S.C. 112, first paragraph, is withdrawn** because Applicant has deleted the phrase "mutants, variants and derivatives of the recombination sites of (a), (b), (c), (d), (e), (f), (g), (h) or (i) which retain the ability to undergo recombination" from the claims.

5. **The prior rejection of Claims 19, 35, 60-79 and 104-128 under 35 U.S.C. 112, second paragraph, are withdrawn** because Applicant has amended the claims to distinguish "a composition comprising" a nucleic acid molecule and at least one topoisomerase. Furthermore, Applicant has deleted the phrase "mutants, variants and derivatives of the recombination sites of (a), (b), (c), (d), (e), (f), (g), (h) or (i) which retain the ability to undergo recombination" from the claims.

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

6. **Claims 20 is rejected under 35 U.S.C. 112, second paragraph**, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claim 20 recites the limitation "said amino acid sequence tag" in reference to the preceding base Claim 19. There is insufficient antecedent basis for this limitation in the claim. The base claims have been amended to replace "amino acid sequence tag" with "SEQ ID NO:6". The Examiner respectfully suggests amending the instantly rejected claim to iterate the same phrase replacement as performed in the base claim. See for example, Claims 61 and 105.

Claim Rejections - 35 USC § 102

7. **The prior rejection of Claims 19, 28, 30-32 and 34-37 under 35 U.S.C. 102(b)** as being anticipated by Hartley et al (AT8) **is withdrawn** in light of Applicant's amendment to base Claim 19 to include the limitation of SEQ ID NO:6 which is not taught by the prior cited art.

8. **The prior rejection of Claims 60, 69-70, 72-73 and 75-79 under 35 U.S.C. 102(b)** as being anticipated by Heyman et al (Genome Research 9: 383-392, 1999) **is withdrawn** in light of Applicant's amendment to base Claim 60 to include the limitation of SEQ ID NO:6 which is not taught by the prior cited art.

9. **The prior rejection of Claims 19, 28, 30-32, 34-38, 60, 69-73, 75-79, 104 and 113-128 are rejected under 35 U.S.C. 102(e)** as being anticipated by Hartley et al (U.S. Patent No. 6,277,608 B1) **is withdrawn** in light of Applicant's amendment to base Claims 19, 60 and 104 to include the limitation of SEQ ID NO:6 which is not taught by the prior cited art.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are

such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

10. **The prior rejection of Claims 19-27, 33, 60-68, 74 and 104-112 under 35 U.S.C. 103(a)** as being unpatentable over Hartley et al (U.S. Patent No. 6,277,608 B1) and Hearn et al (J. Molecular Recognition 14: 323-369, 2001), **is withdrawn** in light of Applicant's amendment to base Claims 19, 60 and 104 to include the limitation of SEQ ID NO:6 which is not taught by the prior cited art.

11. **Claims 19, 30-32, 34-38, 60, 69-73, 75-78, 104, 113-124, 126-128 and 136-138 are rejected under 35 U.S.C. 103(a)** as being unpatentable over as being unpatentable over Hartley et al (U.S. Patent No. 6,277,608 B1; *of record), Hearn et al (J. Molecular Recognition 14: 323-369, 2001; *of record), Cronan et al (AR5; *of record), and Stolz et al (FEBS Letters 440: 213-217, 1998).

The claims are drawn to a method of producing a polynucleotide construct that encodes a fusion protein that comprises the amino acid sequence tag of SEQ ID NO:6, that is a portion of the carboxy terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit capable of being biotinylated.

Hartley et al teach a method for cloning or subcloning desired nucleic acid molecules (col. 20, lines 12-20) into vectors using recombination sites and their corresponding recombination proteins, specifically *attB*, *attP*, *attL*, *attR*, *lox*, *frt* sites, and Cre, Int, IHF, Xis, Fis, Hin, Gin, Cin, and Tn3 resolvase proteins (col.s 22-24) or a topoisomerase, e.g. calf thymus topoisomerase I, which the art recognizes to be a eukaryotic nuclear type IB topoisomerase (col. 8, lines 20-24; col.s 53-54), wherein the invention relates to nucleic acid molecules that may be chimeric and have the "desired characteristic(s) and/or DNA segment(s)" (Abstract), and wherein the product polynucleotides are transformed into a host cell. Examples of desired DNA segments can be, but are not limited to, functional elements, genes or partial genes which encode useful nucleic acids or proteins (column 20, lines 13-20), as exemplified by Example 3 to create fusion proteins comprising an amino acid sequence tag (e.g. glutathione S-transferase, histidine tag) (col. 35).

Hartley et al do not teach the use of *psi*, *dif* or *cer* sites, and the use of their corresponding recombination TndX, XerC or XerD proteins. However, the specification of the instant application discloses that other suitable recombination sites include *psi*, *dif* and *cer* (page 34, [00110]), demonstrating functional equivalency of the claimed recombination sites for the method of producing a polynucleotide construct. Thus, nothing non-obvious is seen with substituting a recombination site taught by Hartley et al with a functionally equivalent *psi*, *dif* or *cer* site, and necessarily TndX, XerC or XerD which recognize said sites, the motivation being that the *psi*, *dif* and *cer* sites increase the arsenal of choices for the artisan to achieve the desired cloning strategy.

Hartley et al do not teach the amino acid sequence tag represented by SEQ ID NO:6 that is capable of being biotinylated; however, at the time of the invention, Hearn et al reviewed the history of peptide fusion handles and affinity cassette methods used in the art to produce and purify recombinant proteins, summarizing the general knowledge and skill in the art for over twenty years. "In all cases, the strategy [of attaching 'affinity tags' or 'peptide handles'] seeks to achieve the most optimal affinity interactions and separation productivity at the laboratory and process scale level" (page 323, column 2). The appropriate and skillful selection of these affinate-affinant systems for a particular family of recombinant proteins forms the basis of the "molecular cassette concept" of high-resolution affinity chromatographic separation of proteins (page 324, column 2). Thus, the art recognizes the modular nature of 'affinity tags' or 'peptide handles' and the routine and common practice to substitute or modify one amino acid tag with another amino acid tag, and even combine multiple amino acid sequence tags, as per the nature and desire of the artisan. Hearn and Acosta teach representative peptide tags, including among others, biotinylation tags (page 334, Table 3).

Hearn et al taught that the number of different tag systems of various molecular sizes, with different binding interactions and affinities, can be selected or designed for fusion to virtually any target protein that can be cloned and expressed in a microbial or eukaryotic host (page 336, column 1, section 5). In particular, Hearn et al make reference to Cronan et al who taught that there are "*obvious advantages*" (emphasis added) of fusing a protein of interest to a protein segment recognized by a coenzyme ligase, such as (i) the fusion proteins could be specifically labeled by growth of cell cultures in the presence of labeled coenzyme; (ii) if the

coenzyme possessed specific binding toward an immobilized ligand, the fusion protein could be readily purified, perhaps in a native form, and (iii) the protein segment fused to the protein of interest would be much smaller than those commonly used, thus giving less alteration of the chemical and biological properties of the protein of interest. Biotin has distinct advantages over the other coenzyme modification groups, the main advantage being the existence of two proteins, avidin and streptavidin, that strongly and specifically bind biotin (Introduction, columns 1-2, joining paragraph). Cronan et al taught the creation of fusion proteins for biotinylation, exemplified using the biotin protein segments derived from the *K. pneumoniae* oxalacetate decarboxylase α subunit.

Neither Hearn et al or Cronan et al taught the amino acid sequence tag of SEQ ID NO:6 that is capable of being biotinylated. However, at the time of the invention, Stolz et al taught the minimal peptide domain of that is a portion of the carboxy terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit capable of being biotinylated, of which SEQ ID NO:6 comprises, necessary for biotinylation (pg 214, Figure 1), wherein the minimal peptide domain begins at amino acid six (Valine 6) of SEQ ID NO:6, and wherein the peptide comprises a five amino acid peptide leader sequence. Stolz et al do not teach the five amino acid leader peptide sequence as present in SEQ ID NO:6; however, absent evidence to the contrary, the nothing non-obvious is seen with replacing the synthetic five amino acid peptide leader of Stolz et al with an endogenous five amino acid leader peptide of SEQ ID NO:6, the motivation being that the five amino acids of SEQ ID NO:6 have short side chains, are relatively unstructured and would provide a peptide "cap" to the conserved tertiary structure created by the C-terminal domain (pg 215, Figure 3) which is biotinylated. Furthermore, the five amino acid peptide leader sequence is part of the wildtype *Klebsiella* oxalacetate decarboxylase α subunit C-terminal domain, and thus does not require the design and synthesis of an artificial peptide leader sequence.

It would have been obvious to one of ordinary skill in the art to modify the polynucleotide construct of Hartley et al that yields a fusion protein with an amino acid tag capable of biotinylation as taught in the instant application with a reasonable chance of success because Hearn et al teach that, for over twenty years, it has been common practice to those of ordinary skill in the art to design and skillfully select peptide fusion handles and affinity cassette

methods. Furthermore, Cronan et al successfully demonstrate the creation of fusion proteins capable of becoming biotinylated using protein segments derived from *K. pneumoniae* oxalacetate decarboxylase α subunit. An artisan would be motivated to use an amino acid sequence tag capable of biotinylation because Cronan et al teach that the extremely specific and high affinity binding of biotin by avidin and streptavidin results in specific detection systems of very high sensitivity, which may also be used to purify the biotinylated proteins (page 10330, columns 1-2, joining paragraph) and it has been common practice to those of ordinary skill in the art to design and skillfully select peptide fusion handles and affinity cassette methods to achieve optimal production and purification of recombinant proteins. Thus, epitope-tagging heterologous proteins via post-translational biotinylation is a well-recognized approach in the art.

Thus, the invention as a whole is *prima facie* obvious.

12. **Claims 19-21, 33, 60-62, 74 and 104-106 are rejected under 35 U.S.C. 103(a)** as being unpatentable over Hartley et al (U.S. Patent No. 6,277,608 B1; *of record), Hearn et al (J. Molecular Recognition 14: 323-369, 2001; *of record), Cronan et al (AR5; *of record), and Stolz et al (FEBS Letters 440: 213-217, 1998), as applied to Claims 19, 60 and 104 above, and in further view of Arienne et al (AR1; *of record), as applied respectively to the limitations of biotinylation and enterokinase protease.

The claims are drawn to a method of producing a polynucleotide construct that encodes a fusion protein that comprises the amino acid sequence tag of SEQ ID NO:6, that is a portion of the carboxy terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit capable of being biotinylated, and an amino acid sequence that is capable of being cleaved by one or more proteases—the preferred embodiment being enterokinase.

Hartley et al do not explicitly teach the amino acid sequence tag be capable of post-translational modification, specifically biotinylation, nor that the polynucleotide construct also encode an amino acid sequence capable of being cleaved by a protease, specifically enterokinase. However, at the time of the invention, Hearn et al reviewed the history of peptide fusion handles and affinity cassette methods, alone or in combination with amino acid sequences capable of being cleaved by a protease, used in the art to produce and purify recombinant proteins,

summarizing the general knowledge and skill in the art for over twenty years. "In all cases, the strategy [of attaching 'affinity tags' or 'peptide handles'] seeks to achieve the most optimal affinity interactions and separation productivity at the laboratory and process scale level" (page 323, column 2). Hearn et al also summarized the use of endopeptidases, including among others, enterokinase (page 332, Table 1), commonly used in the art at the time of filing of the instant application to cleave one's protein of interest from the affinity tag (page 334, Table 3) and facilitate protein purification. For example, Hearn et al reference the PinPoint™ system (Promega, USA), in which the product polynucleotide encodes a fusion protein that contains an amino acid sequence tag capable of biotinylation, in combination with an endoprotease cleavage site, e.g. Factor Xa (page 351, columns 1-2, joining paragraph).

It would have been obvious to one of ordinary skill in the art to modify the polynucleotide construct of Hartley et al that yields a fusion protein to also comprise an amino acid sequence for protease cleavage as taught in the instant application with a reasonable chance of success because Hearn et al had summarized the common practice in the art regarding the successful use of 'affinity tags' in combination with amino acid sequences capable of being cleaved by a protease, e.g. enterokinase, as recited in the instant application. Furthermore, Airenne et al demonstrated the successful method of peptide tagging (e.g. avidin) a polypeptide of interest (e.g. Hevein) with a proteolytic cleavage site for Factor Xa, enterokinase, thrombin and tobacco etch virus to cleave Hevein from the amino acid tag and facilitate isolation and purification of the Hevein polypeptide (page 143, c1), thus demonstrating the art-recognized functional equivalence of the utilizing any of these four proteases to cleave one's protein of interest from the amino acid sequence tag. An artisan would be motivated to include an amino acid sequence capable of being cleaved by a protease when creating a fusion protein comprising one's protein of interest and a biotinylated amino acid sequence tag because Cronan et al teaches that the extremely tight binding of biotin and biotinylated proteins to avidin or streptavidin cannot be reversed by elution with biotin, and thus elution of bound proteins requires much harsher methods (page 10330, columns 1-2, joining paragraph). Cronan et al suggest that one approach to address this challenge is to engineer a site for protease cleavage (e.g. the PinPoint™ system, Promega, USA). Such a fusion protein could be bound to immobilized avidin or streptavidin, washed free of contaminating proteins, and simultaneously eluted in native form

(and freed of the biotinylated segment) by treatment of the column matrix with the designated protease. Avidin and streptavidin are extremely insensitive to proteases in general, and since the amino acid sequences of both proteins are known, selection of proteases unable to cleave avidin and streptavidin is readily accomplished. Furthermore, Airenne et al teach that protease cleavage sites can easily be attached to the fusion protein by adding the appropriate sequences for cloning.

Thus, the invention as a whole is *prima facie* obvious.

13. **Claims 60, 76-79, 104 and 122-125 are rejected under 35 U.S.C. 103(a)** as being unpatentable over Hartley et al (U.S. Patent No. 6,277,608 B1; *of record), Hearn and Acosta (J. Molecular Recognition 14: 323-369, 2001; *of record), Cronan et al (AR5), Stolz et al (FEBS Letters 440: 213-217, 1998), as applied to Claims 60, 76-78, 104 and 122-124 above, and in further view of Heyman et al (Genome Research 9: 383-392, 1999; *of record).

The prior cited art does not teach the use of a poxvirus topoisomerase; however, at the time of the invention, Heyman et al taught methods of cloning one's gene of interest into expression vectors using topoisomerases, wherein the topoisomerase is the vaccinia virus-encoded eukaryotic type I topoisomerase (page 383, Introduction, column 1), resulting in fusion proteins comprising an amino acid sequence tag (page 385, Figure 2).

It would have been obvious to one of ordinary skill in the art to substitute the topoisomerase I of Hartley et al with the topoisomerase I of Heyman et al with a reasonable chance of success because, absent evidence to the contrary, both enzymes are type IB topoisomerases and thus functionally equivalent. An artisan would be motivated to substitute the calf thymus topoisomerase I with the vaccinia topoisomerase I because Heyman et al teach that the vaccinia topoisomerase I has a unique activity to both cleave and re-join DNA strands with high sequence specificity (pg 383, col. 1, ¶1).

Thus, the invention as a whole is *prima facie* obvious.

Conclusion

14. No claim is allowed.

Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Kevin K. Hill, Ph.D. whose telephone number is 571-272-8036. The examiner can normally be reached on Monday through Friday, between 9:00am-6:00pm EST.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Joseph T. Woitach can be reached on 571-272-0739. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

Kevin K. Hill

Joseph Woitach
AU1633